REMARKS

Claims 1, 3-8, 21 and 35-38 are pending. No new matter has been added by way of the present amendments. Claim 1 has been amended to modify the language 'the genus related to *Imperata*, etc' as supported by the present specification. This does not raise new issues since the Examiner has already considered these limitations. In the event that the present submission does not place the application into condition for allowance, entry thereof is respectfully requested as placing the application into better form for appeal.

In view of the following remarks, Applicants respectfully request that the Examiner reconsider and withdraw all rejections.

Issues under 35 U.S.C §112, first paragraph

The Examiner has maintained the rejection of claims 1, 3-8, 21 and 35-38 under 35 U.S.C §112, first paragraph for alleged lack of written description of the invention as explained at pages 2-9 of the outstanding final Office Action. Applicants respectfully traverse.

1. Written Description Issue

The Examiner has asserted that according to the instant disclosure, Applicants were only in possession of a 67 kDa protein that was isolated from *Imperata cylindrical* and not from that other genuses such as *Lolim, Phleum* and *Cynodon*. Therefore, The Examiner asserts that the specification also fails to provide adequate written description for claimed protein isolated from *Lolium perenne* (hereinafter "*Lp*"), *Phleum pratense* (hereinafter "*Pp*") or *Cynodon datylon* (hereinafter "*Cd*"). As mentioned in arguments of records, Applicants respectfully disagree with the Examiner.

First, it is clear that the present specification provides adequate written description as to isolation of pollen grains from the above grasses. Applicants respectfully direct the Examiner to review the present specification page 2, lines 33-35, page 8, lines 11-14, page 9, lines 5-30 and the Examples. In particular, page 9, lines 5-25 discloses a process of purification of the novel 67 kDa protein capable of inhibiting anthrax toxin activity by using of steps (a) – (f). Among them, steps (a) and (b) are directed to the isolation of the protein by the protein extraction from the buffer (a) and the purification by column chromatography (b). Also, these two isolation steps disclose specified conditions such as the name of the buffer, suspending time, speed of the centrifuge, and purifying method. Further, these two steps correspond to Example 2 (step (a)), Example 3 (step (a)) and Example 4 (step (b) and Fig. 1). The present specification, page 8, lines 10-15 and page 9, lines 25-30 clearly discloses that the pollen grains for purification of the protein in step (a) are collected from grasses selected from group comprising of *Ic, Lp, Pp, Cd* and related genus. Accordingly, by using these steps, such proteins are isolated from *Lp, Pp* or *Cd*. Therefore, Applicants respectfully submit that since the present specification provides sufficient and adequate written description of how to isolate the protein from *Lp, Pp* and *Cd* based on page 9 and the Examples, one skilled in the art would conclude that Applicants were in possession of a 67 kDa protein from these three grasses as well as *Ic*.

Second, it is evident that Figure 3 supports the isolation of the protein from the above grasses. A Western Blot (alternately, immunoblot) is a method to detect a specific protein in a given sample of tissue extract. Herein, in the present application, the Western blotting was performed on the isolated 67 kDa proteins from *Ic*. A similar protein was demonstrated to exist in *Lp*, *Pp* and *Cd* using patients' sera specific to these grasses according to the above mentioned

steps (a)-(d). Showing reactivity by very sensitive and specific procedure, the Western blot method proves that the 67 kDa type protein is present in these grass extracts. This data provides that 67-kDa protein or similar kind of protein is present in these grasses and it can be isolated from these four grasses. Fig 3 also reveals that allergenic cross-reactivity and immunoblotting, which are sensitive techniques, have established the presence of a similar kind of protein in these grasses. Further, *Imperata* and *Cynodon* are considered as tropical grasses. These grasses have more than one common epitope because the blot has been done with a polyclonal antibody. Therefore, Applicants respectfully traverse the Examiner's assertion that at most *Cd*, *Lp* and *Pp* have at least one common epitope.

Third, it is improper that the Examiner requires Applicants limit their claims to subject matter that has actually been reduced to practice. It is not the law in the United States that an Applicant must restrict his claims to working examples actually reduced to practice. *See, e.g. In re Rasmussen*, 221 USPQ 323 (CCPA 1981); *In re Koller, Hartl & Kirchner*, 204 USPQ 702 (CCPA 1980). Also working examples are not at all required for a disclosure to adequately support a claim. *In re Strahilevitz*, 212 USPQ 561 (CCPA 1982. In order to meet the written description requirement of 35 U.S.C. §112, first paragraph, a patent specification must contain a written description to "allow persons of ordinary skill in the art to recognize that [the inventor] invented what is claimed." *Gentry Gallery, Inc. v. Berkline Corp.*, 45 USPQ2d 1498, 1503 (Fed. Cir. 1998) (quoting In re Gosteli, 10 USPQ 2d 1614, 1618 (Fed. Cir. 1989).

In view of the above, Applicants respectfully submit that those of skill in the art would understand that Applicants were in possession of the subject matter claimed at the time of filing because the present specification pages (e.g., 2, 3, 6, 8, 9, and 11) describe the claimed invention

in sufficient detail that one skilled in the art can reasonably conclude that the inventors had possession of the claimed invention.

In summary, the present specification serves to adequately support the present breadth of the claims. Therefore, Applicants respectfully reconsideration and withdrawal of the instant rejection

2. Enablement Issue

The Examiner has also asserted that the instant specification has failed to correlate *in vivo* treatment of anthrax using the claimed protein and the *in vitro* treatment using the claimed protein. Applicants respectfully disagree with the Examiner.

First, the Applicant's burden in writing the specification is merely to establish, by a preponderance of the evidence, not beyond reasonable doubt or even clear and convincing evidence, that the protein in question would have the utility asserted and so that the use of the invention is enabled. To this end, the present specification quite clearly demonstrates that the protein of the invention is able to inhibit anthrax toxin *in vitro*. This *in vitro* test is performed on a tissue culture experiment that is similar to *in vivo* treatment and can be extrapolated thereto because once 67 kDa protein blocks the entry of anthrax toxin, a higher number of cells survives against toxin exposure by this treatment. This *in vitro* experiment was done in a dose dependent manner to prove that specific receptor blocked the entry of lethal factor into the cells and that made cell survival. Therefore, the fact that the viability of cells is quantitatively measured on an *in vitro* test means that the 67 kDa protein is effective in reducing the cleavage of anthrax toxin protective antigen thereby reducing the entry of lethal factor and finally enhancing cell survival.

Second, the Examiner has questioned, "how would the claimed 67 kDa protein react when administered *in vivo* to patients that produce high levels of IgE neutralizing antibodies due to allergic reactions?" Applicants respectfully submit that the Examiner is confusing 'the activity of the protein' and 'IgE binding'. The claimed protein binds to IgE. IgE binding is a property of the immune system to recognize as an allergen. Nobody knows why a protein behaves as an allergen. The other property of the 67 kDa protein is the activity of the protein. The activity of the protein is directed to inhibiting the cleavage of the protective antigen thereby blocking the entry of lethal factor. The 67 kDa protein is also an allergen and treatment of allergies thereto can be performed by immunotherapy using this protein. Immunotherapy is a process similar to vaccination. Hence, the blocking of antibodies generated by this protein will protect individuals from allergic reactions to grasses. Applicants respectfully request that the Examiner remember the two properties of the same protein (IgE binding and the protease inhibitor activity).

Third, the Examiner has questioned 'preincubation' as to the *in vivo* treatment. Applicants respectfully submit that the function of 67 kDa protein is to inhibit the cleavage of anthrax toxin. To inhibit the cleavage of anthrax toxin protective antigen, before the start of the experiment, the cells were preincubated with the 67 kDa protein. The protein also works in a similar manner if the protein is added instantaneously, but preincubation may give better results. An experiment has already been conducted to prove and activity of the preincubated or not-preincubated remains similar, and literature in the art also supports these findings. Such will be effective *in vivo* because *Bacillus anthracis* secrets the toxin components individually and these toxin component, protective antigen will not be functional, until cleaved. On cleavage it binds to

lethal factor and then the complex enters the cell to show activity. In a similar manner, Applicants have conducted an experiment where they have incubated both protective antigen and 67 kDa protein together. 67 kDa have inhibited anthrax toxin activity. Therefore, the claimed protein can likely retain its activity in circulation, and is thus effective in inhibiting anthrax toxicity to mammalian cells.

Fourth, the 67 kDa protein is not a "protective antigen." Protective antigen is the name of the anthrax toxin protein, which on cleavage allows lethal factor or edema factor to bind and thereby these proteins enter the cells to give biochemical activity. Applicants again emphasize that the 67 kDa protein inhibits the cleavage of anthrax toxin protective antigen thereby the enzymatic proteins (LF or EF) are not delivered to the cytosol of the cells. The 67-kDa protein does not give protective immunity but it inhibits the cleavage of protective antigen (a protein of anthrax toxin).

Fifth, the administration of proteins as antitoxins is well-known in the art. The claimed protein can be administered by inhalation, as it will interact directly with the affected cells because lethal factor action is reported in respiratory system. The specification provides an indication of the concentration of the protein of the invention effective in inhibiting anthrax toxin, and is it within the skill of the practitioner of pharmacology to perform known pharmacodynamic experiments based on this information to arrive at an effective *in vivo* dose.

Based on the above, an *in vitro* test may be sufficient to establish a therapeutic use for a novel compound. Therefore, Applicants request reconsideration and withdrawal of this rejection.

Issues under 35. U.S.C. 112, second paragraph

The Examiner has maintained the rejection of claims 1, 3-8, 21 and 35-38 under 35 U.S.C. 112, second paragraph for the reasons recited at page 15 of the outstanding final Office Action.

Without acquiescing to the Examiner's allegations made to reject the claims and to expedite prosecution only, claim 1 has been amended to entirely modify the language of (ix) to read "the 67 kDa protein is isolated from *Imperata cylindrica* and demonstrated that a similar protein is present in *Lolium perenne, Phleum pratense* and *Cynodon dactylon*."

Second, the Examiner has rejected claim 6 asserting that the phrase "partially inhibits" is unclear. Applicants respectfully submit that "partially inhibits" mean that cleavage of protective antigen was inhibited in a dose dependent manner and at this concentration it does not inhibit completely. However, Applicants already deleted the word "partially" from the claim 6 on submission of Reply dated October 25, 2006.

In view of the above, Applicants respectfully submit that the present claims fully meet the requirement of 35 U.S.C. §112, second paragraph. Accordingly, the Examiner is respectfully requested to reconsider and withdraw this rejections.

Issues under 35 U.S.C § 102(a)

The Examiner has rejected claims 1, 3-8, 21 and 35-38 under 35 U.S.C §102(a) as being anticipated by Bijli et al (Clin. Exp. Allergy, January 2003) (hereinafter "Bijli 2003"). Applicants respectfully traverse.

In the paper of Bijli 2003 (page 65), the protein was not isolated but only observed with SDS-PAGE. As well known in the art, SDS-PAGE is a technique to visualize the separated proteins by Coomassie, silver staining, etc. Bijli's proteins were not isolated by this SDS-PAGE technique because the protein was not purified.

Bijli 2003 (Fig. 2) shows that there were 37 different numbers of proteins. Accordingly, during the SDS-PAGE and Western blotting, some of the proteins reacted with the antibodies. But there was no disclosure of purification of protein in this paper and therefore the activity of the purified protein cannot be determined and has not been presented in this paper by Bijli 2003. Also, the paper describes that after keeping the *Imperata* extracts with different stabilizers, protein could maintain the allergenic activity in the extract. It means the protein may be a mixture of proteins or degradation products of high molecular weight proteins. Therefore, Applicants respectfully submit that the protein seen in the Bijli 2003 paper is not the same as that of the present invention because visualization of protein in a mixture may not be sufficient for determining the activity. Accordingly, the Examiner's requirement for "side by side" comparison and presentation of evidence in declaration form is not necessary. Also, the Examiner may consult a biochemist/biotechnologist for a clear picture on this issue. The Examiner may present examples where an SDS-PAGE is used for protein isolation and determining activity. Only after the protein is isolated, then activity of the protein can be determined. Here it would be worth to mention that one skilled in the art cannot isolate the protein by SDS-PAGE but only visualize the proteins.

To support this, the present protein was purified by a hydrophobic column chromatography as defined in claim 37. Claim 37, depending from claim 1, recites how the

protein is isolated. Claim 1 also clearly defines the protein as an "isolated" form. Therefore, the "isolated" protein of the present invention cannot be regarded the same protein of the Bijli 2003 which is not isolated and may be a mixture of proteins or degradation products of high molecular weight proteins. Accordingly, the Examiner is respectfully requested to reconsider and withdraw this anticipation rejection.

Issues under 35 U.S.C. §102 (b)

The Examiner has rejected claims 1, 3-8, 21 and 35-38 under 35 U.S.C §102(b) as being anticipated by Bijli et al (Journal of Immunological Methods 260 (Feb. 2002, 91-96) (hereinafter referred to as "Bijli 2002"). Applicants respectfully traverse with substantially the same argument as the above §102(a).

A protein cannot be isolated without a purification process. In this context, Bijli 2002 (page 92-93) does not teach that a 67 kDa protein is isolated from *Imperata*. This protein was just extracted from a standard SDS-PAGE method. This extract may be a mixture of proteins or degradation products. This protein did not go through a purification process and it was not isolated. Thus, the activity of the protein cannot be determined and compared with that of the present invention. Bijli 2002 does not provide teachings regarding the biological activity of the claimed protein. Accordingly, it cannot be judged that protein with the same molecular weight have same activity and property. Applicants have given examples already about proteins having same molecular weight and with different properties in a various extracts

This paper argues that a protein(s) or degraded protein of higher molecular weight separates around the same molecular weight. However, the protein was never isolated and not

confirmed to be the same. The protein extracted from pollen of *Imperata* under room temperature and the 67 kDa protein of the present invention may not provide any evidence of similarity in these proteins because the extracts will contain mixture of many proteins unlike a purified and isolated protein of the present invention. Accordingly, the Examiner's requirement for "side by side" comparison and presentation of evidence in declaration form is not necessary.

To support the above, the present protein was purified by a hydrophobic column chromatography as defined in claim 37. Claim 37, depending from claim 1, recites how the protein is isolated. Claim 1 also clearly defines the protein as an "isolated" form. Therefore, the 'isolated' protein of the present invention cannot be regarded the same protein of the Bijli 2002 which is not isolated and may be a mixture of proteins or degradation products of high molecular weight proteins. Accordingly, the Examiner is respectfully requested that reconsideration and withdrawal of this anticipation rejection.

The Examiner has rejected claims 1, 3-8, 21 and 35-38 under 35 U.S.C §102 (b) as being anticipated by Verma et al (International Archives of Allergy and Immunology, 2000, 122:251-256)(hereinafter referred to as "Verma"). Applicants respectfully traverse.

The protein isolated by Verma is different from that isolated in the present application. Applicants have used a different protocol than Verma for purification of the 67 kDa protein. It is supported by Verma's claim that this protein cannot be sequenced because the N-terminal is blocked i.e. glycosylated. But the 67 kDa protein in the present invention was sequenced using Edman degradation. This shows that the property of both proteins are different and they are not the same proteins. A side-by-side comparison of the purified protein (Verma) submitted on

February 6, 2006. The protein has different molecular weight on SDS-PAGE visualization as shown in Declaration.

Verma has purified the protein by different column chromatography such as Ion exchange, Gel Filtration, Liquid (unlike the hydrophobic interaction used in the present invention). Verma's protein is a protein and also mentioned in the paper as "homogenous peak and a single band (page 254)." Again the protein's N-terminal is blocked and sequencing was not possible or this protein and therefore further cleaved with CNBr to characterize it further. However, the protein described in the present invention could be sequenced and therefore both proteins are different. Claim 37 in the present application reflects that isolated protein is obtained by hydrophobic column chromatography. As mentioned above, it does not mean that protein with the same molecular weight have same activity. Applicants have given examples already that the protein with same molecular weight from the same extract has different properties as shown in the Declaration. Therefore, both proteins are different each other in activity. Reconsideration and withdrawal of this rejection are therefore respectfully requested.

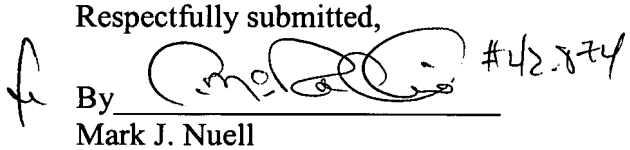
In view of the above, Applicants respectfully submit that the present claims define allowable subject matter. Accordingly, the Examiner is respectfully requested to withdraw all rejections and allow the currently pending claims

If the Examiner has any questions or comments, please contact the undersigned at the offices of Birch, Stewart, Kolasch & Birch, LLP.

If necessary, the Commissioner is hereby authorized in this, concurrent, and future replies, to charge payment or credit any overpayment to our Deposit Account No. 02-2448 for any additional fees required under 37 C.F.R. § 1.16 or under § 1.17; particularly, extension of time fees.

Dated: July 2, 2007

Respectfully submitted,

By  #42,874

Mark J. Nuell
Registration No.: 36,623
BIRCH, STEWART, KOLASCH & BIRCH, LLP
8110 Gatehouse Road
Suite 100 East
P.O. Box 747
Falls Church, Virginia 22040-0747
(703) 205-8000
Attorney for Applicant